

WEEK13

July 18

- STREAKING of plates for Orientation.
- 2 plates prepared by Promoter.

July 19

- Due to no positive result. We started again
- Transformation: nrfA and sqr biobricks into Dh5 α
- Followed by plating

July 20 digestion

For enzyme test

- **EcoRI:** Control DNA is **λ DNA (N3011G)**
- **XbaI:** Control DNA is **λ DNA (dam-/HindIII digest) (N3012G)**
- **PstI:** Control DNA is **λ DNA (N3011G)**
- **T4 DNA Ligase:** Control DNA is **λ DNA (dam-/HindIII digest) (N3012G)**

Control reaction conditions:

- NE Buffer - 4 μ l
- Control DNA - 1 μ g (2 μ l)
- Nuclease Free Water- 33 μ l (total reaction volume is 40 μ l)
- Enzyme - 1 μ l

Incubate at 37 $^{\circ}$ C for 1-2 hours.

Heat inactivate the enzyme at 65 $^{\circ}$ C for 20 min.

Run at 80-90 V in a 1% gel.

GEL RUN

Order:

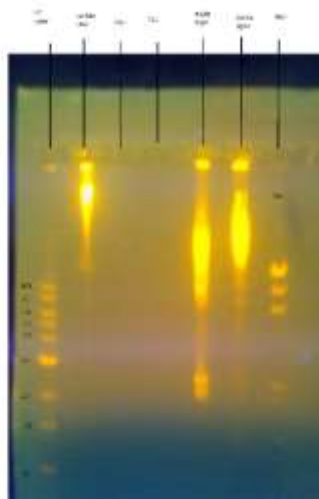
Name	Ladder(1kb)	Lambda DNA	E	P	Hind III Digest	L	X
Volume(ul)	4	2	10	10	2	10	10

- Dye 1 ul...

Tubes named as:

Ecor1	Pst1	Xba1	T4 DNA Ligase
E	P	X	L

GEL RESULT:



INNOCULATION of Transformed cells::

Names of test tubes as:

- NrfA Kana
- SQR Kana

July 22

Plasmid prep of:

- NrfA Kana
- SQR Kana

GEL RUN:

Result plasmids bends observed.

July 23

DIGESTION of both the isolated plasmids:

	NrfA	SQR
Tube Name	NrfA	Sqr
Water	21	21
Plasmid	2	2
Neb buffer 2.1	5	5
Ecor1	1	1
Pst1	1	1